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=> s tpa or t-pa of (plasminogen (w) activator)
MISSING OPERATOR 'OF (PLASMINOGE'
The search profile that was entered contains terms or
nested terms that are not separated by a logical operator.

=> s tpa or t-pa or (plasminogen (w) activator)
L1      166707 TPA OR T-PA OR (PLASMINOGEN (W) ACTIVATOR)

=> s l1 and (k429? or h417? or d477?)
L2            6 L1 AND (K429? OR H417? OR D477?)

=> dup rem l2
PROCESSING COMPLETED FOR L2
L3            3 DUP REM L2 (3 DUPLICATES REMOVED)

=> d py l3 1-3

L3   ANSWER 1 OF 3  CAPLUS  COPYRIGHT 2002 ACS
PY   1999
     1999

L3   ANSWER 2 OF 3  CAPLUS  COPYRIGHT 2002 ACS
PY   1998
     1998
     1998
     2001
     1999
     2000
     2001

L3   ANSWER 3 OF 3  BIOSIS  COPYRIGHT 2002 BIOLOGICAL ABSTRACTS INC.DUPLICATE 1
PY   1990


=> d 3 l3 ab so ti py au

L3   ANSWER 3 OF 3  BIOSIS  COPYRIGHT 2002 BIOLOGICAL ABSTRACTS INC.DUPLICATE 1
AB   In contrast to most other serine proteases, tissue-type
       ***plasminogen***     ***activator***   (  ***t***  -  ***PA***  )
     possesses enzymatic activity as the one-chain zymogen form. The hypothesis
     that lysine residues 277 or 416 may be involved in stabilization of an
     active conformation of one-chain   ***t***  -  ***PA***   via salt-bridge
     formation with aspartic acid residue 477 was tested by site-directed
     mutagenesis. Four recombinant   ***t***  -  ***PA***   mutants were
     constructed. The amidolytic activities of these analogues were compared to
     that of authentic    ***t***  -  ***PA***  . Substitution of arginine-275
     provided an analogue ([R275G]  ***t***  -  ***PA***  ) resistant to
     plasmin cleavage. The amidolytic activity of [R275G]  ***t***  -  ***PA***
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was comparable to that of authentic one-chain ***t*** - ***PA*** , and so was the activity of [R275L,K277L] ***t*** - ***PA*** , in which additional substitution of lysine residue 277 was carried out. This suggested that its presence was nonessential for obtaining one-chain ***t*** - ***PA*** activity. In contrast, substitution of lysine residue 416 to obtain [K416S] ***t*** - ***PA*** and [K416S, ***H417T***] ***t*** - ***PA*** resulted in substantial quenching of amidolytic one-chain activity. As expected, the amidolytic activities of the two-chain forms were less affected by the substitution. Involvement of lysine residue 416 in one-chain ***t*** - ***PA*** activity was also indicated by decreased activities of [K416S] ***t*** - ***PA*** and [K416S, ***H417T***] ***t*** - ***PA*** with plasminogen as the substrate. The one-chain activity of the lysine residue 416 substitution analogues was partially restored in the presence of fibrin. This could indicate that strong ligands such as fibrin might provide an alternative stabilization of the active conformation of one-chain ***t*** - ***PA*** .
SO BIOCHEMISTRY, (1990) 29 (14), 3451-3457.
CODEN: BICHAW. ISSN: 0006-2960.
TI QUENCHING OF THE AMIDOLYTIC ACTIVITY OF ONE-CHAIN TISSUE-TYPE ***PLASMINOGEN*** ***ACTIVATOR*** BY MUTATION OF LYSINE-416.
PY 1990
AU PETERSEN L C; BOEL E; JOHANNESSEN M; FOSTER D

=> d hist

(FILE 'HOME' ENTERED AT 10:53:16 ON 05 SEP 2002)

FILE 'BIOSIS, MEDLINE, CAPLUS, EMBASE, SCISEARCH' ENTERED AT 10:53:47 ON 05 SEP 2002
L1 166707 S TPA OR T-PA OR (PLASMINOGEN (W) ACTIVATOR)
L2 6 S L1 AND (K429? OR H417? OR D477?)
L3 3 DUP REM L2 (3 DUPLICATES REMOVED)

=> s l1 and zymogen
L4 997 L1 AND ZYMOGEN

=> dup rem l4
PROCESSING COMPLETED FOR L4
L5 364 DUP REM L4 (633 DUPLICATES REMOVED)

=> s l5 and fibrin
L6 97 L5 AND FIBRIN

=> s l6 and py<=1996
1 FILES SEARCHED...
4 FILES SEARCHED...
L7 62 L6 AND PY<=1996

=> s l7 and (275 or 276)
L8 2 L7 AND (275 OR 276)

=> d ab l8

L8 ANSWER 1 OF 2 BIOSIS COPYRIGHT 2002 BIOLOGICAL ABSTRACTS INC.
AB In contrast to most other serine proteases, tissue-type ***plasminogen*** ***activator*** (***t*** - ***PA***) possesses enzymatic activity as the one-chain ***zymogen*** form. The hypothesis that lysine residues 277 or 416 may be involved in stabilization of an active conformation of one-chain ***t*** - ***PA*** via salt-bridge formation with aspartic acid residue 477 was tested by site-directed mutagenesis. Four recombinant ***t*** - ***PA*** mutants were constructed. The amidolytic activities of these analogues were compared to that of authentic ***t*** - ***PA*** . Substitution of arginine- ***275*** provided an analogue ([R275G] ***t*** - ***PA***) resistant to plasmin cleavage. The amidolytic activity of [R275G] ***t*** - ***PA*** was comparable to that of authentic one-chain ***t*** - ***PA*** , and so was the activity of [R275L,K277L] ***t*** - ***PA*** , in which additional substitution of lysine residue 277 was carried out. This suggested that its presence was nonessential for obtaining one-chain ***t*** - ***PA*** activity. In contrast, substitution of lysine residue 416 to obtain [K416S] ***t*** - ***PA*** and [K416S,H417T] ***t*** - ***PA*** resulted in substantial quenching of amidolytic one-chain activity. As expected, the amidolytic activities of the two-chain forms were less affected by the

substitution. Involvement of lysine residue 416 in one-chain ***t*** - ***PA*** activity was also indicated by decreased activities of [K416S] ***t*** - ***PA*** and [K416S,H417T] ***t*** - ***PA*** with plasminogen as the substrate. The one-chain activity of the lysine residue 416 substitution analogues was partially restored in the presence of ***fibrin*** . This could indicate that strong ligands such as ***fibrin*** might provide an alternative stabilization of the active conformation of one-chain ***t*** - ***PA*** .

=> d ab l8 2

L8 ANSWER 2 OF 2 BIOSIS COPYRIGHT 2002 BIOLOGICAL ABSTRACTS INC.
AB Activation of the ***zymogen*** form of a serine protease is associated with a conformational change that follows proteolysis at a specific site. Tissue-type ***plasminogen*** ***activator*** (***t*** - ***PA***) is homologous to mammalian serine proteases and contains an apparent activation cleavage site at arginine- ***275*** . To clarify the functional consequences of cleavage at arginine- ***275*** of ***t*** - ***PA*** , site-specific mutagenesis was performed to convert arginine- ***275*** to a glutamic acid. The mutant enzyme (designated Arg- ***275*** .fwdarw. Glu ***t*** - ***PA***) could be converted to the two-chain form by Staphylococcus aureus V8 protease but not by plasmin. The one-chain form was 8 times less active against the tripeptide substrate H-D-isoleucyl-L-prolyl-L-arginine-p-nitroanilide (S-2288), and the ability of the enzyme to activate plasminogen in the absence of fibrinogen was reduced 20-50 times compared to the two-chain form. In contrast, one-chain Arg- ***275*** .fwdarw. Glu ***t*** - ***PA*** has equal activity to the two-chain form when assayed in the presence of physiological levels of fibrinogen and plasminogen. ***Fibrin*** bound significantly more of the one-chain form of ***t*** - ***PA*** than the two-chain form for both the wild-type and mutated enzymes. One- and two-chain forms of the wild-type and mutated ***plasminogen*** ***activators*** slowly formed complexes with plasma protease inhibitors, although the one-chain forms showed decreased complex formation with .alpha.2-macroglobulin. The one-chain form of ***t*** - ***PA*** therefore is fully functional under physiologic conditions and has an increased ***fibrin*** binding compared to the two-chain form.

=> d ab l8 2 py so ti au

L8 ANSWER 2 OF 2 BIOSIS COPYRIGHT 2002 BIOLOGICAL ABSTRACTS INC.
AB Activation of the ***zymogen*** form of a serine protease is associated with a conformational change that follows proteolysis at a specific site. Tissue-type ***plasminogen*** ***activator*** (***t*** - ***PA***) is homologous to mammalian serine proteases and contains an apparent activation cleavage site at arginine- ***275*** . To clarify the functional consequences of cleavage at arginine- ***275*** of ***t*** - ***PA*** , site-specific mutagenesis was performed to convert arginine- ***275*** to a glutamic acid. The mutant enzyme (designated Arg- ***275*** .fwdarw. Glu ***t*** - ***PA***) could be converted to the two-chain form by Staphylococcus aureus V8 protease but not by plasmin. The one-chain form was 8 times less active against the tripeptide substrate H-D-isoleucyl-L-prolyl-L-arginine-p-nitroanilide (S-2288), and the ability of the enzyme to activate plasminogen in the absence of fibrinogen was reduced 20-50 times compared to the two-chain form. In contrast, one-chain Arg- ***275*** .fwdarw. Glu ***t*** - ***PA*** has equal activity to the two-chain form when assayed in the presence of physiological levels of fibrinogen and plasminogen. ***Fibrin*** bound significantly more of the one-chain form of ***t*** - ***PA*** than the two-chain form for both the wild-type and mutated enzymes. One- and two-chain forms of the wild-type and mutated ***plasminogen*** ***activators*** slowly formed complexes with plasma protease inhibitors, although the one-chain forms showed decreased complex formation with .alpha.2-macroglobulin. The one-chain form of ***t*** - ***PA*** therefore is fully functional under physiologic conditions and has an increased ***fibrin*** binding compared to the two-chain form.
PY 1987
SO BIOCHEMISTRY, (1987) 26 (2), 338-343.
CODEN: BICHAW. ISSN: 0006-2960.
TI FUNCTIONAL ROLE OF PROTEOLYTIC CLEAVAGE AT ARGININE- ***275*** OF HUMAN TISSUE ***PLASMINOGEN*** ***ACTIVATOR*** AS ASSESSED BY SITE-DIRECTED MUTAGENESIS.

AU TATE K M; HIGGINS D L; HOLMES W E; WINKLER M E; HEYNEKER H L; VEHAR G A

=> d hist

(FILE 'HOME' ENTERED AT 10:53:16 ON 05 SEP 2002)

FILE 'BIOSIS, MEDLINE, CAPLUS, EMBASE, SCISEARCH' ENTERED AT 10:53:47 ON 05 SEP 2002

L1 166707 S TPA OR T-PA OR (PLASMINOGEN (W) ACTIVATOR)
L2 6 S L1 AND (K429? OR H417? OR D477?)
L3 3 DUP REM L2 (3 DUPLICATES REMOVED)
L4 997 S L1 AND ZYMOGEN
L5 364 DUP REM L4 (633 DUPLICATES REMOVED)
L6 97 S L5 AND FIBRIN
L7 62 S L6 AND PY<=1996
L8 2 S L7 AND (275 OR 276)

=> s l5 and salt bridge
L9 7 L5 AND SALT BRIDGE

=> s l5 and (salt (w) bridge)
L10 7 L5 AND (SALT (W) BRIDGE)

=> s l1 and (salt (w) bridge)
L11 60 L1 AND (SALT (W) BRIDGE)

=> dup rem l11
PROCESSING COMPLETED FOR L11
L12 21 DUP REM L11 (39 DUPLICATES REMOVED)

=> s l12 and py<=1996
1 FILES SEARCHED...
4 FILES SEARCHED...
L13 8 L12 AND PY<=1996

=> d l13 ab au ti py so

L13 ANSWER 1 OF 8 BIOSIS COPYRIGHT 2002 BIOLOGICAL ABSTRACTS INC.
AB Trypsinogen is converted to trypsin by the removal of a peptide from the N terminus, which permits formation of a ***salt*** ***bridge*** between the new N-terminal Ile (residue 16) and Asp194. Formation of this ***salt*** ***bridge*** triggers a conformational change in the "activation domain" of trypsin, creating the S1 binding site and oxyanion hole. Thus, the activation of trypsinogen appears to represent an example of protein folding driven by electrostatic interactions. The following trypsin mutants have been constructed to explore this problem: Asp194Asn, Ile16Val, Ile16Ala, and Ile16Gly. The bovine pancreatic trypsin inhibitor (BPTI), benzamidine, and leupeptin affinities and activity and pH-rate profiles of these mutants have been measured. The changes in BPTI and benzamidine affinity measure destabilization of the activation domain. These experiments indicate that hydrophobic interactions of the Ile16 side chain provide 5 kcal/mol of stabilization energy to the activation domain while the ***salt*** ***bridge*** accounts for 3 kcal/mol. Thus, hydrophobic interactions provide the majority of stabilization energy for the trypsinogen to trypsin conversion. The pH-rate profiles of I16A and I16G are significantly different than the pH-rate profile of trypsin, further confirming that the activation domain has been destabilized. Moreover, these mutations decrease k-cat/K-m and leupeptin affinity in parallel with the decrease in stability of the activation domain. Acylation is selectively decreased, while substrate binding and deacylation are not affected. Together these observations indicate that the stability of protein structure is an important component of transition state stabilization in enzyme catalysis. These results also suggest that active zymogens can be created without providing a counterion for Asp194, and thus have important implications for the elucidation of the structural features which account for the zymogen activity of tissue ***plasminogen*** ***activator*** and urokinase.
AU Hedstrom, Lizbeth (1); Lin, Tiao-Yin; Fast, Walter
TI Hydrophobic interactions control zymogen activation in the trypsin family of serine proteases.
PY 1996
SO Biochemistry, (1996) Vol. 35, No. 14, pp. 4515-4523.
ISSN: 0006-2960.

=> d l13 ab au ti py so 2-8

L13 ANSWER 2 OF 8 BIOSIS COPYRIGHT 2002 BIOLOGICAL ABSTRACTS INC.
AB Tissue-type ***plasminogen*** ***activator*** (***t*** - ***PA***), a multidomainal serine proteinase of the trypsin-family catalyses the rate-limiting step in fibrinolysis, the activation of plasminogen to the fibrin-degrading proteinase plasmin. Trigonal crystals have been obtained of the recombinant catalytic domain of human two-chain ***t*** - ***PA*** , consisting of a 17 residue A chain and the 252 residue B chain. Its X-ray crystal structure has been solved applying Patterson and isomorphous replacement methods and has been crystallographically refined to an R-value of 0.184 at 2.3 ANG resolution. The chain fold, active-site geometry and Ile276-Asp477 ***salt*** ***bridge*** are similar to that observed for trypsin. A few surface-located insertion loops differ significantly however. The disulfide bridge Cys315-Cys384, practically unique to the ***plasminogen*** ***activators*** , is incorporated without drastic conformational changes as the insertion loop preceding Cys384 makes a bulge on the molecular surface. The unique basic insertion loop Lys296-Arg304 flanking the primed subsites, which has been shown to be of importance for PAI-1 binding and for fibrin specificity, is partially disordered; it can therefore freely adapt to proteins docking to the active site. The S1 pocket of ***t*** - ***PA*** is almost identical to that of trypsin, whereas the S2 site is considerably reduced in size by the imposing Tyr368 side-chain, in agreement with the measured preference for P1 Arg and P2 Gly residues. The neighbouring S3-S4 hydrophobic groove is mainly hydrophobic in nature. The structure of the proteinase domain of two-chain ***t*** - ***PA*** suggests that the formation of a ***salt*** ***bridge*** between Lys429 and Asp477 may contribute to the unusually high catalytic activity of single-chain ***t*** - ***PA*** , thus stabilizing the catalytically active conformation without unmasking the Ile276 amino terminus. Modeling studies show that the covalently bound kringle 2 domain in full-length ***t*** - ***PA*** could interact with an extended hydrophobic groove in the catalytic domain; in such a docking geometry its "lysine binding site" and the "fibrin binding patch" of the catalytic domain are in close proximity.
AU Lamba, Doriano; Bauer, Margit; Huber, Robert; Fischer, Stephen; Rudolph, Rainer; Kohnert, Ulrich; Bode, Wolfram (1)
TI The 2.3 A crystal structure of the catalytic domain of recombinant two-chain human tissue-type ***plasminogen*** ***activator*** .
PY 1996
SO Journal of Molecular Biology, (1996) Vol. 258, No. 1, pp. 117-135. ISSN: 0022-2836.

L13 ANSWER 3 OF 8 BIOSIS COPYRIGHT 2002 BIOLOGICAL ABSTRACTS INC.
AB Prothrombin fragment 2 (the second kringle) has been co-crystallized with PPACK (D-Phe-Pro-Arg)-thrombin and the structure of the non-covalent complex has been determined and refined (R = 0.16) at 3.2 ANG resolution using X-ray crystallographic methods. The kringles interact with thrombin at a site that has previously been proposed to be the heparin binding region. The latter is a highly electropositive surface near the C-terminal helix of thrombin abundant in arginine and lysine residues. These form ***salt*** ***bridges*** with acidic side chains of kringle 2. Somewhat unexpectedly, the negative groups of the kringle correspond to an enlarged anionic center of the lysine binding site of lysine binding kringles such as plasminogen K1 and K4 and ***TPA*** K2. The anionic motif is DGDEE in prothrombin kringle 2. The corresponding cationic center of the lysine binding site region has an unfavorable Arg71Phe substitution but Lys35 is conserved. However, the folding of fragment 2 is different from that of prothrombin kringle 1 and other kringles: the second outer loop possesses a distorted two-turn helix and the hairpin beta-turn of the second inner loop pivots at V64 and D70 by 60 degree . The Lys35 is located on a turn of the helix, which causes it to project into solvent space in the fragment 2-thrombin complex, thereby devastating the cationic center of the lysine binding site. Since fragment 2 has not been reported to bind lysine, it most likely has a different inherent folding conformation for the second outer loop, while the movement of the V64-D70 beta-turn is most likely a conformational change accompanying complexation, both of which reveal a new heretofore unsuspected flexibility in kringles. The fragment 2-thrombin complex is the first cassette module-catalytic domain interaction determined for a multi-domain blood protein and only the second domain-domain interaction to be described among such proteins, the other being Ca+2 prothrombin fragment 1 (Gla-domain and kringle 1). It has been possible to propose a reasonable model for the four domain prothrombin structure using these two double domain structures.

AU Arni, Raghuvir K.; Padma[illegible]han, Kaillathe; Padmanabhan, K[illegible].; Wu, Tswei-Ping; Tulinsky, A.[illegible])
TI Structure of the non-covalent complex of prothrombin kringle 2 with PPACK-thrombin.
PY 1994
SO Chemistry and Physics of Lipids, (1994) Vol. 67-68, No. 0, pp. 59-66. ISSN: 0009-3084.

L13 ANSWER 4 OF 8 BIOSIS COPYRIGHT 2002 BIOLOGICAL ABSTRACTS INC.
AB Both human and bovine prothrombin fragment 2 (the second kringle) have been cocrystallized separately with human PPACK (D-Phe-Pro-Arg)-thrombin, and the structures of these noncovalent complexes have been determined and refined (R = 0.155 and 0.157, respectively) at 3.3- ANG resolution using X-ray crystallographic methods. The kringles interact with thrombin at a site that has previously been proposed to be the heparin binding region. The latter is a highly electropositive surface near the C-terminal helix of thrombin abundant in arginine and lysine residues. These form ***salt*** ***bridges*** with acidic side chains of kringle 2. Somewhat unexpectedly, the negative groups of the kringle correspond to an enlarged anionic center of the lysine binding site of lysine binding kringles such as plasminogens K1 and K4 and ***TPA*** K2. The anionic motif is DGDEE in prothrombin kringle 2. The corresponding cationic center of the lysine binding site region has an unfavorable Arg70Asp substitution, but Lys 35 is conserved. However, the folding of fragment 2 is different from that of prothrombin kringle 1 and other kringles: the second outer loop possesses a distorted two-turn helix, and the hairpin beta-turn of the second inner loop pivots at Val64 and Asp70 by 60 degree . Lys35 is located on a turn of the helix, which causes it to project into solvent space in the fragment 2-thrombin complex, thereby devastating any vestige of the cationic center of the lysine binding site. Since fragment 2 has not been reported to bind lysine, it most likely has a different inherent folding conformation for the second outer loop, as has also been observed to be the case with ***TPA*** K2 and the urokinase kringle. The movement of the Val64-Asp70 beta-turn is most likely a conformational change accompanying complexation, which reveals a new heretofore unsuspected flexibility in kringles. The fragment 2-thrombin complex is only the second cassette module-catalytic domain structure to be determined for a multidomain blood protein and only the third domain-domain interaction to be described among such proteins, the others being factor Xa without a Gla domain and Ca-2+ prothrombin fragment 1 with a Gla domain and a kringle.
AU Arni, Raghuvir K.; Padmanabhan, Kaillathe; Padmanabhan, K. P.; Wu, Tswei-Ping; Tulinsky, A. (1)
TI Structures of the noncovalent complexes of human and bovine prothrombin fragment 2 with human PPACK-thrombin.
PY 1993
SO Biochemistry, (1993) Vol. 32, No. 18, pp. 4727-4737. ISSN: 0006-2960.

L13 ANSWER 5 OF 8 BIOSIS COPYRIGHT 2002 BIOLOGICAL ABSTRACTS INC.
AB In contrast to most other serine proteases, tissue-type ***plasminogen*** ***activator*** (***t*** - ***PA***) possesses enzymatic activity as the one-chain zymogen form. The hypothesis that lysine residues 277 or 416 may be involved in stabilization of an active conformation of one-chain ***t*** - ***PA*** via ***salt*** - ***bridge*** formation with aspartic acid residue 477 was tested by site-directed mutagenesis. Four recombinant ***t*** - ***PA*** mutants were constructed. The amidolytic activities of these analogues were compared to that of authentic ***t*** - ***PA*** . Substitution of arginine-275 provided an analogue ([R275G] ***t*** - ***PA***) resistant to plasmin cleavage. The amidolytic activity of [R275G] ***t*** - ***PA*** was comparable to that of authentic one-chain ***t*** - ***PA*** , and so was the activity of [R275L,K277L] ***t*** - ***PA*** , in which additional substitution of lysine residue 277 was carried out. This suggested that its presence was nonessential for obtaining one-chain ***t*** - ***PA*** activity. In contrast, substitution of lysine residue 416 to obtain [K416S] ***t*** - ***PA*** and [K416S,H417T] ***t*** - ***PA*** resulted in substantial quenching of amidolytic one-chain activity. As expected, the amidolytic activities of the two-chain forms were less affected by the substitution. Involvement of lysine residue 416 in one-chain ***t*** - ***PA*** activity was also indicated by decreased activities of [K416S] ***t*** - ***PA*** and [K416S,H417T] ***t*** - ***PA*** with plasminogen as the substrate. The one-chain activity of the lysine residue 416 substitution analogues was partially restored in the presence of fibrin. This could indicate that strong ligands such as fibrin might provide an alternative

stabilization of the act[redacted] conformation of one-chain *[redacted]*** -
PA .

AU PETERSEN L C; BOEL E; JOHANNESSEN M; FOSTER D
TI QUENCHING OF THE AMIDOLYTIC ACTIVITY OF ONE-CHAIN TISSUE-TYPE
PLASMINOGEN ***ACTIVATOR*** BY MUTATION OF LYSINE-416.
PY 1990
SO BIOCHEMISTRY, (1990) 29 (14), 3451-3457.
CODEN: BICHAW. ISSN: 0006-2960.

L13 ANSWER 6 OF 8 MEDLINE
AB The contribution of intersubunit interactions to allosterically induced conformational changes in phosphorylase are considered. Phosphorylase a, Pa (phosphorylated at Ser-14), is significantly in the active (R) conformation, while phosphorylase b, Pb (nonphosphorylated), is predominantly in the inactive (T) conformation. The structure of glucose-inhibited (***T***) ***Pa*** has been determined at 2.5-A resolution and atomic coordinates have been measured. These data have been used to calculate the solvent accessible surface area at the subunit interface and map noncovalent interactions between protomers. The subunit contact involves only 6% of the Pa monomer surface, but withdraws an area of 4,600 A2 from solvent. The contact region is confined to the N-terminal (regulatory) domain of the subunit. Half of the residues involved are among the 70 N-terminal peptides. A total of approximately 100 atoms take part in polar or nonpolar contacts of less than 4.0 A with atoms of the symmetry-related monomer. The contact surface surrounds a central cavity at the core of the interface of sufficient volume to accommodate 150-180 solvent molecules. There are four intersubunit ***salt*** ***bridges*** . Two of these (Arg 10/Asp 32, Ser-14-P/Arg 43) are interactions between the N-terminus of one protomer with an alpha-helix loop segment near the N-terminus of the symmetry-related molecule. These two are relatively solvent accessible. The remainder (Arg 49/Glu 195, Arg 184/Asp 251) are nearer the interface core and are less accessible. The ***salt*** ***bridges*** at the N-terminus are surrounded by the polar and nonpolar contacts which may contribute to their stability. Analysis of the difference electron density between the isomorphous Pa and Pb crystal structures reveals that the N-terminal 17 residues of Pb are disordered. Pb thus lacks two intermolecular and one intersubunit (Ser-14-P/Arg 69) salt linkage present in Pa. The absence of these interactions in Pb is manifested in the difference in the free energy of T leads to R activation, which is 4 kcal more than that for Pa. Difference Fourier analysis of the T leads to R transition in substrate-activated crystals of Pa suggests that the 70 N-terminal residues undergo a concerted shift towards the molecular core; ***salt*** ***bridges*** are probably conserved in the transition. It is proposed that the N-terminus, when "activated" by phosphorylation (via a specific kinase) behaves as an intramolecular "effector" of the R state in phosphorylase and serves as the vehicle of homotropic cooperativity between subunits of the dimer.
AU Sprang S; Fletterick R J
TI Subunit interactions and the allosteric response in phosphorylase.
PY 1980
SO BIOPHYSICAL JOURNAL, ***(1980 Oct)*** 32 (1) 175-92.
Journal code: 0370626. ISSN: 0006-3495.

L13 ANSWER 7 OF 8 CAPLUS COPYRIGHT 2002 ACS
AB Assocn. energies of 3 porphyrins bearing meso-4-pyridinium, -4-anilinium, or -4-benzoate substituents (TPPyP, ***TPA*** , TCP) are evaluated in water with 23 different ligands. UV titrns. show excellent fit and const. values (usually .+-.10%) for the equil. consts. K evaluated at different wavelengths as well as isosbestic points, securing 1:1 complexes as well as negligible self-assocn. of the porphyrins. Complexation-induced proton NMR shifts (CIS) show shielding of the ligands by up to -8.0 ppm and of up to -3.4 ppm on the porphyrins with arom. ligands; they agree with tight face-to-face conformations. Ionic binding contributions can be factorized with 5.2 .+-. 1.1 kJ per mol and per ***salt*** ***bridge*** (or ion pair). After subtraction of the ***salt*** ***bridge*** increments, const. binding free energies are obsd. with .DELTA.Gvdw = 7.2 .+-. 1.5 kJ mol-1 for all benzene derivs. and .DELTA.Gvdw = 15.8 .+-. 1.8 kJ mol-1 for all naphthalene-like, and .DELTA.Gvdw = 18.5 .+-. 0.5 kJ mol-1 for phenanthrene-like derivs. Deviations are obsd. with bulky substituents like +NMe3 or SO3- groups which allow no close contact between the arom. planes, as evident from CHARMm simulations. Heterocyclic electroneutral ligands show the same const. .DELTA.Gvdw values, independent of no. and position of the heteroatoms. Comparison of 33 different .DELTA.Gvdw contributions with the no. of double bonds occurring in the ligands yields, for the first time, a comprehensive

description of stacking interactions with an increment of 4 .+-. 0.15 kJ per mol and .pi.-electron Measurements with satd. ligand of comparable surface show no .DELTA.Gvdw contributions, indicating that the stacking is not driven by solvophobic but by dispersive forces. Copper(II) in the porphyrins (CuTPPyP) leads within .+-.0.4 kJ mol-1 to the same assocn. energies as without metal, whereas introduction of zinc leads to a decrease by usually 3.4 kJ mol-1 . Axial orientations of the ligands are obsd. with complexes of .alpha.,.omega.-diaminoalkanes with a gable or sandwich Zn-porphyrin dimer. The .DELTA.Gcplx values, measured in chloroform, increase with the match between the Zn and the N atoms. Measurements of K in binary methanol-water mixts. with four complexes (e.g. TPPyP + phenanthroline) show linear correlations with solvent polarity or hydrophobicity parameters. Linear correlations are also found for the first time between the complexation-induced Soret band wavelength changes and the corresponding .DELTA.Gcplx values.
AU Schneider, Hans-Joerg; Wang, Manxue
TI Supramolecular Chemistry. 49. Ligand-Porphyrin Complexes: Quantitative Evaluation of Stacking and Ionic Contributions
PY ***1994***
SO Journal of Organic Chemistry (***1994***), 59(24), 7464-72
CODEN: JOCEAH; ISSN: 0022-3263

L13 ANSWER 8 OF 8 EMBASE COPYRIGHT 2002 ELSEVIER SCI. B.V.
AB Protein C inhibitor (PCI) inhibits multiple plasma serine proteases. To determine which residues contribute to its specificity of inhibition, 19 mutations in the reactive site loop of PCI (from Thr352 to Arg357) were generated and assayed with thrombin, activated protein C (APC), and factor Xa. To identify the intermolecular interactions responsible for these kinetics, a molecular model of PCI was generated using .alpha.1-protease inhibitor and ovalbumin as templates. This model of PCI was docked with thrombin, followed by extensive energy minimization, to determine a lowest energy complex. The resulting docked complex was used as a template to form molecular models of PCI-APC and PCI-factor Xa complexes. The best inhibitors of thrombin contained Pro or Gly at the P2 position in place of Phe353, with 2- and 7-fold increases in activity, respectively. These substitutions reduced steric interactions with the 60-insertion loop unique to thrombin. The best inhibitors of APC and factor Xa contained Arg at the P3 position in place of Thr352, with 2- and 5-fold increases in inhibition rates, respectively. The molecular model predicts that Arg in this position could form a ***salt*** ***bridge*** with Glu217 of each protease. Changing Arg357 at the P3' position had little effect on protease inhibition, consistent with the observation in the model that this residue points toward the body of PCI, forming a ***salt*** ***bridge*** with Glu220. Given its broad specificity of inhibition, PCI has proven very useful in understanding the nature of serpin- protease interactions using multiple mutations in a serpin assayed with multiple proteases.
AU Cooper S.T.; Whinna H.C.; Jackson T.P.; Boyd J.M.; Church F.C.
TI Intermolecular interactions between protein C inhibitor and coagulation proteases.
PY ***1995***
SO Biochemistry, (1995) 34/40 (12991-12997).
ISSN: 0006-2960 CODEN: BICHAW

=> d hist

(FILE 'HOME' ENTERED AT 10:53:16 ON 05 SEP 2002)

FILE 'BIOSIS, MEDLINE, CAPLUS, EMBASE, SCISEARCH' ENTERED AT 10:53:47 ON 05 SEP 2002
L1 166707 S TPA OR T-PA OR (PLASMINOGEN (W) ACTIVATOR)
L2 6 S L1 AND (K429? OR H417? OR D477?)
L3 3 DUP REM L2 (3 DUPLICATES REMOVED)
L4 997 S L1 AND ZYMOGEN
L5 364 DUP REM L4 (633 DUPLICATES REMOVED)
L6 97 S L5 AND FIBRIN
L7 62 S L6 AND PY<=1996
L8 2 S L7 AND (275 OR 276)
L9 7 S L5 AND SALT BRIDGE
L10 7 S L5 AND (SALT (W) BRIDGE)
L11 60 S L1 AND (SALT (W) BRIDGE)
L12 21 DUP REM L11 (39 DUPLICATES REMOVED)
L13 8 S L12 AND PY<=1996

=>

---Logging off of STN---

=>
Executing the logoff script...

=> LOG Y

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FULL ESTIMATED COST	81.11	81.32

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